



INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁷ : C07K 14/34, C12P 21/02, C12N 15/31, A61P 35/00 // C12R 1:16		A1	(11) International Publication Number: WO 00/50449 (43) International Publication Date: 31 August 2000 (31.08.00)																					
(21) International Application Number: PCT/GB00/00680		Halldis [NO/NO]; Nycomed Imaging AS, Nycoveien 1-2, N-0401 Oslo (NO).																						
(22) International Filing Date: 25 February 2000 (25.02.00)		(74) Agents: GRANT, Anne, Rosemary et al.; Frank B. Dehn & Co., 179 Queen Victoria Street, London EC4V 4EL (GB).																						
(30) Priority Data: 9904582.5 26 February 1999 (26.02.99) GB		(81) Designated States: AE, AL, AM, AT, AT (Utility model), AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, CZ (Utility model), DE, DE (Utility model), DK, DK (Utility model), DM, EE, EE (Utility model), ES, FI, FI (Utility model), GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KR (Utility model), KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SK (Utility model), SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).																						
(71) Applicant (for all designated States except US): NYCOMED IMAGING AS [NO/NO]; Nycoveien 1-2, N-0401 Oslo (NO).		Published With international search report. Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.																						
(72) Inventors; and (75) Inventors/Applicants (for US only): WOLFE, Henry, Raphael [US/US]; Nycomed Amersham Imaging Inc., 101 Carnegie Centre, Princeton, NJ 08540 (US). MERCHANT, Fahar [CA/CA]; Intelligene Expressions, Inc., Edmonton Research Park, 138-94 Street, Edmonton, Alberta T6N 1J3 (CA). MERCHANT, Rosamina [CA/CA]; Intelligene Expressions, Inc., Edmonton Research Park, 138-94 Street, Edmonton, Alberta T6N 1J3 (CA). BLACK, Christopher, D., V. [GB/US]; Nycomed Amersham Imaging Inc., 101 Carnegie Centre, Princeton, NJ 08540 (US). STORFLOR, Harry [NO/NO]; Nycomed Imaging AS, Nycoveien 1-2, N-0401 Oslo (NO). STOKKE, Geir, O. [NO/NO]; Nycomed Imaging AS, Nycoveien 1-2, N-0401 Oslo (NO). HELLEBUST,																								
(54) Title: PROCESS FOR PRODUCTION DIPHTHERIA TOXIN																								
<p style="text-align: center;">PRODUCTION OF CRM107 BY <i>C. DIPHTHERIAE</i> GROWN IN A 10-L FERMENTER IN NIH MEDIUM CONTAINING 1.5% GLUCOSE. LOT D + CYSTEINE AND 2% YE. LOT E + CYSTINE AND 1% YE. (SDS-PAGE)</p> <table border="1"> <caption>Estimated data from the graph</caption> <thead> <tr> <th>Time (h)</th> <th>1.5% glucose + cystine (mg/L)</th> <th>1.5% glucose + cysteine (mg/L)</th> </tr> </thead> <tbody> <tr><td>0</td><td>0</td><td>0</td></tr> <tr><td>9</td><td>0</td><td>15</td></tr> <tr><td>10</td><td>22</td><td>25</td></tr> <tr><td>11</td><td>95</td><td>38</td></tr> <tr><td>12</td><td>85</td><td>10</td></tr> <tr><td>13</td><td>98</td><td>0</td></tr> </tbody> </table>				Time (h)	1.5% glucose + cystine (mg/L)	1.5% glucose + cysteine (mg/L)	0	0	0	9	0	15	10	22	25	11	95	38	12	85	10	13	98	0
Time (h)	1.5% glucose + cystine (mg/L)	1.5% glucose + cysteine (mg/L)																						
0	0	0																						
9	0	15																						
10	22	25																						
11	95	38																						
12	85	10																						
13	98	0																						
(57) Abstract																								
<p>The present invention provides a method of purifying diphtheria toxin comprising (1) fermenting a microorganism strain capable of producing diphtheria toxin using glucose as a carbon source, said method comprising adding glucose to a growing culture whereby the addition of glucose maintains microorganism growth effective to support diphtheria toxin production; and (2) purifying the diphtheria toxin from the culture by contacting a toxin containing preparation derived therefrom with an ion exchange matrix, eluting a fraction containing the toxin, applying the eluate to a hydrophobic matrix, and eluting a fraction containing the toxin.</p>																								

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece			TR	Turkey
BG	Bulgaria	HU	Hungary	ML	Mali	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MN	Mongolia	UA	Ukraine
BR	Brazil	IL	Israel	MR	Mauritania	UG	Uganda
BY	Belarus	IS	Iceland	MW	Malawi	US	United States of America
CA	Canada	IT	Italy	MX	Mexico	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NE	Niger	VN	Viet Nam
CG	Congo	KE	Kenya	NL	Netherlands	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NO	Norway	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	NZ	New Zealand		
CM	Cameroon			PL	Poland		
CN	China	KR	Republic of Korea	PT	Portugal		
CU	Cuba	KZ	Kazakhstan	RO	Romania		
CZ	Czech Republic	LC	Saint Lucia	RU	Russian Federation		
DE	Germany	LI	Liechtenstein	SD	Sudan		
DK	Denmark	LK	Sri Lanka	SE	Sweden		
EE	Estonia	LR	Liberia	SG	Singapore		